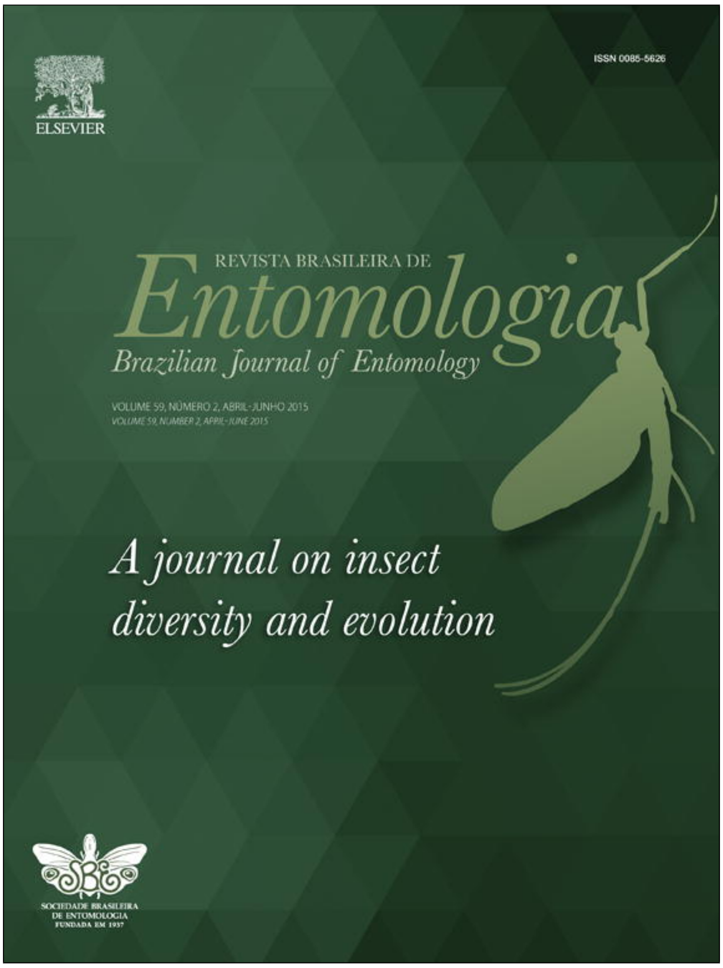
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Short Communication

Sex ratios in juveniles and adults of *Dichroplus maculipennis* (Blanchard) and *Borellia bruneri* (Rehn) (Orthoptera: Acrididae)

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ABSTRACT

Dichroplus maculipennis and *Borellia bruneri* are two of the 18 grasshopper species of actual or potential economic relevance as pests in Argentina. The objective of this study was to estimate the sex ratios for adults and older nymphs of *D. maculipennis* and *B. bruneri* in the field, and analyze possible temporal variations. The study was conducted during seven seasons (2005–06 to 2011–12) in representative plant communities of the southern Pampas region. A total of 4536 individuals of *D. maculipennis*, and 6038 individuals of *B. bruneri* were collected. The sex ratio registered in older nymphs for *D. maculipennis* and *B. bruneri* did not deviate from a 1:1 ratio ($p > 0.05$), suggesting that these species have such a primary sex ratio. However, a significant bias in sex composition in adults of both species was observed ($p < 0.05$). The sex ratio in adults of *D. maculipennis* was different in five of the 18 sampling dates carried out. In three sampling dates it was biased toward males, while in the other two it was biased toward females. Taking into account the sex ratio by sampling season, significant differences were recorded in two seasons. In 2007–08 the sex ratio was biased toward males (1 F:2.26 M), while in 2008–09 it was biased toward females (1.35 F:1 M). The sex ratio in adults of *B. bruneri* was always biased toward males ($p < 0.05$). We conclude that results obtained in this study indicate that various factors like differential survival, dispersion, predation, among others, could have modified the primary sex ratio in these species.

Populations are characterized by structural attributes, such as density, spatial distribution, dispersion pattern, age, genetic composition, and sex ratio. These attributes change through time and space as a result of responses to changing environmental conditions (Schowalter, 2006). Several authors have highlighted the importance of estimating the sex ratio of a population in studies on life cycles, population dynamics, sexual selection, and also as a key factor in biological control programs (Mulkern, 1983; Smith et al., 2004).

Dichroplus maculipennis (Blanchard, 1851) and *Borellia bruneri* (Rehn, 1906) are two of the 18 grasshopper species considered to be of actual or potential economic relevance as pests in Argentina (Cigliano et al., 2014). *D. maculipennis* is one of the most damaging pests mainly in areas of the Pampas and Patagonia regions (Lange et al., 2005). *B. bruneri* is also a common species of grasshopper communities inhabiting the Pampas grasslands and one of the most harmful in the Pampas of Uruguay (COPR, 1982). Both are univoltine with obligatory embryonic diapause (Mariottini et al., 2011a), and they have a wide geographic distribution, occurring in southernmost Brazil, much of Argentina, Chile, and Uruguay (Eades et al., 2013).

Since late 2005 grasshopper monitoring is being conducted in representative plant communities of the southern Pampas, an area affected by grasshopper pests. Major outbreaks of *D. maculipennis* and *B. bruneri* were recorded from late 2008 to early 2011 causing significant economic losses, affecting a wide variety of crops and natural grasslands in an area of approximately 2.5 million ha (Mariottini et al., 2012). Considering the importance of knowing the structural attributes of populations of pest species, the objective was to estimate the sex ratios for adults and older nymphs (juveniles) of *D. maculipennis* and *B. bruneri* in the field and analyze possible temporal fluctuations.

The study was conducted in Laprida county (36° 02′S, 59° 06′ W), Buenos Aires province, in the South of the Pampas region as defined by Cabrera and Willink (1973). Grasslands are the dominant vegetation type in this region, farming and livestock production being widespread (Batista et al., 2005). Grasshopper samplings were performed from late spring to late summer during seven consecutive seasons (2005–06 to 2011–12). The sampling sites represent the most common plant communities in the area, classified into four

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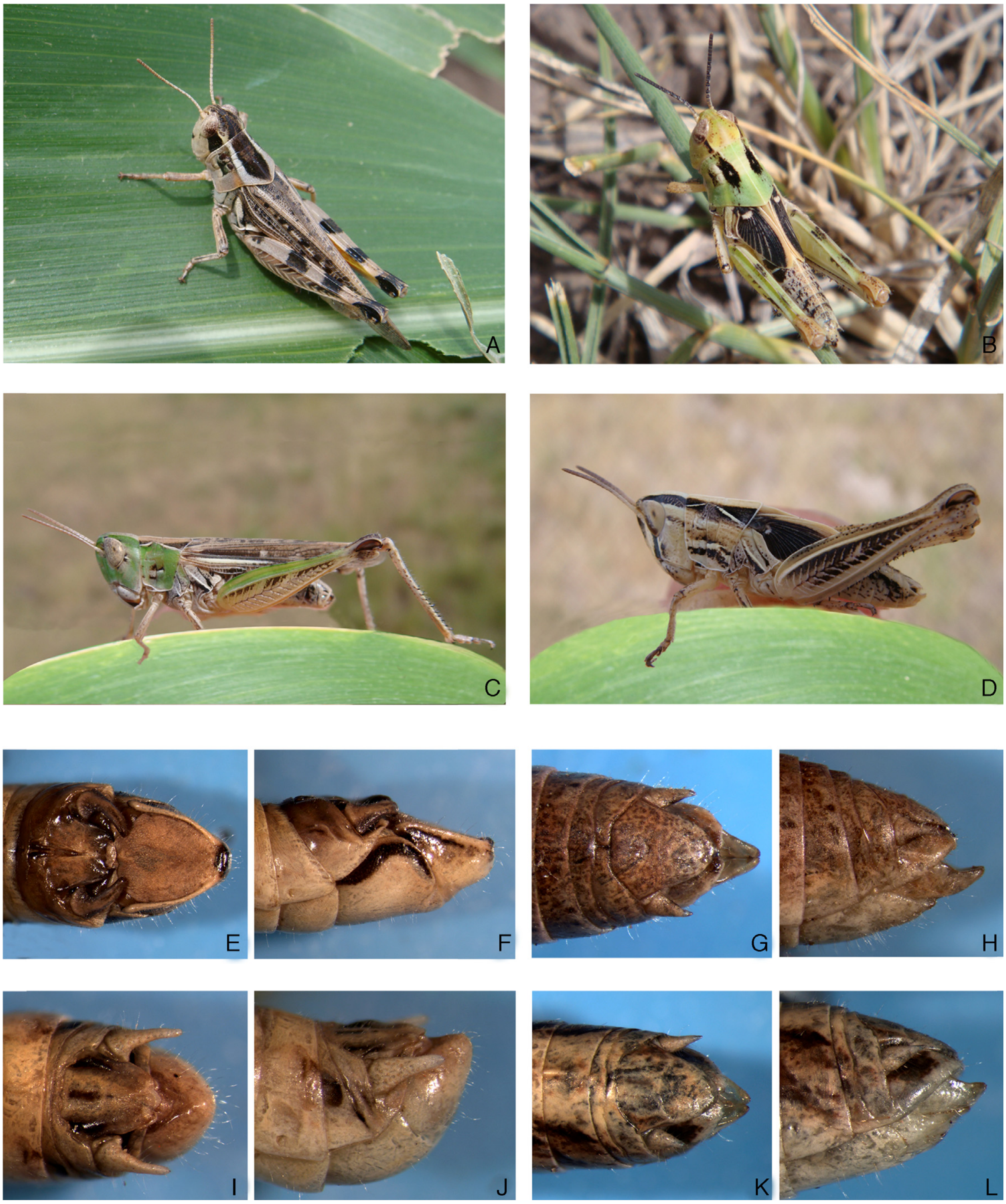

Fig. 1. (A), (B), (E)–(H) *Dichroplus maculipennis*. (A) Adult female. (B) Juvenile female. (E) and (F) Male external genitalia, dorsal (E) and lateral (F) views. (G) and (H) Juvenile female external genitalia, dorsal (G) and lateral (H) views. (C), (D), (I)–(L) *Borellia bruneri*. (C) Adult female. (D) Juvenile female. (I) and (J) Male external genitalia, dorsal (I) and lateral (J) views. (K) and (L) Juvenile female external genitalia, dorsal (K) and lateral (L) views.

categories: native grasslands, disturbed grasslands, implanted pastures, and halophilous grasslands (Mariottini et al., 2013). Three replicates of each category were established, giving a total of 12 sites. Grasshoppers were collected by 200 sweeps of entomological nets (diameter: 40 cm, depth: 75 cm, arc of sweep: 180°) along transects through the vegetation at each site, an efficient method for obtaining representative samples of grasshopper communities (Larson et al., 1999). Grasshoppers collected were taken to the laboratory for identification following Carbonell et al. (2006). Sex ratios were determined in older nymphs (instars four and five in *B. bruneri*, and four to six in *D. maculipennis*) and adults of both species by examination of the external characteristics of genitalia (Uvarov, 1966) (Fig. 1). Sex ratios were estimated for adults and nymphs independently, considering the total of adults or nymphs collected in all sites at each collecting date. Differences between the proportion of males and females of each species were analyzed by a test of comparison of two proportions, and sex ratios were compared by sampling time and for sampling season. The software Xlstat 7.5.3 was used (Addinsoft, 2005).

A total of 4536 individuals of *D. maculipennis* (2974 adults, 1562 nymphs) and 6038 individuals of *B. bruneri* (5327 adults, 711 nymphs) were collected. Differences in the sex ratio of

Table 1
Sex ratios in older nymphs and adults of *Dichroplus maculipennis* in Laprida county, Buenos Aires, Argentina, during 2005–2012.

Season	Date	Stage	Number		Ratio		
			F	M	F	M	p
2005–06	01/10/06	A	8	7	0.53	0.47	0.79
	02/16/06	A	5	6	0.45	0.55	0.76
	Total		13	13	0.5	0.5	1
2006–07	12/12/06	–	–	–	–	–	–
	12/24/06	A	10	18	0.36	0.64	0.15
	01/10/07	A	22	22	0.5	0.5	1.00
	02/05/07	A	10	9	0.53	0.47	0.82
	Total		42	49	0.46	0.54	0.57
2007–08	12/31/07	A	18	28	0.42	0.58	0.29
	01/28/08	A	39	65	0.37	0.63	0.10
	02/23/08	A	8	41	0.16	0.84	<0.0001
	Total		65	134	0.33	0.67	<0.0001
2008–09	12/13/08	N	115	121	0.49	0.51	0.69
		A	127	129	0.50	0.50	0.87
	01/03/09	A	226	132	0.63	0.37	<0.0001
	Total		468	382	0.55	0.45	0.003
2009–10	11/27/09	N	429	476	0.47	0.53	0.12
	12/14/09	N	134	112	0.54	0.46	0.16
		A	40	65	0.38	0.62	0.02
	01/03/10	A	97	182	0.35	0.65	<0.0001
	01/28/10	A	291	210	0.58	0.42	0.00
	Total		991	1045	0.49	0.51	0.232
2010–11	12/11/10	A	50	66	0.43	0.57	0.14
	01/17/11	A	403	414	0.49	0.51	0.57
	Total		453	480	0.49	0.51	0.38
2011–12	12/06/11	N	95	80	0.54	0.46	0.25
		A	3	2	0.60	0.40	0.66
	01/04/12	A	105	116	0.48	0.52	055
	Total		203	198	0.48	0.52	0.46

A, adult; F, female; M, male; N, nymph; p, test result of comparison of two proportions.

Table 2
Sex ratios in older nymphs and adults of *Borellia bruneri* in Laprida county, Buenos Aires, Argentina, during 2005–2012.

Season	Date	Stage	Number		Ratio		
			F	M	F	M	p
2005–06	01/10/06	A	61	150	0.29	0.71	<0.0001
	02/16/06	A	34	59	0.37	0.63	0.015
	Total		95	209	0.31	0.69	<0.0001
2006–07	12/12/06	N	51	43	0.54	0.46	0.41
	12/24/06	A	41	120	0.25	0.75	<0.0001
	01/10/07	A	159	434	0.27	0.73	<0.0001
	02/05/07	A	95	146	0.39	0.61	0.001
	Total		346	743	0.32	0.68	<0.0001
2007–08	12/31/07	A	140	377	0.27	0.73	<0.0001
	01/28/08	A	320	611	0.34	0.66	<0.0001
	02/23/08	A	105	311	0.25	0.75	<0.0001
	Total		565	1299	0.30	0.70	<0.0001
2008–09	12/13/08	N	126	109	0.54	0.46	0.27
		A	30	66	0.31	0.69	<0.0001
	01/03/09	A	618	836	0.43	0.57	<0.0001
	Total		774	1011	0.43	0.57	<0.0001
2009–10	11/27/09	N	43	41	0.51	0.49	0.72
	12/14/09	N	152	146	0.51	0.49	0.83
		A	23	57	0.29	0.71	0.001
	01/03/10	A	101	180	0.36	0.64	<0.0001
	01/28/10	A	55	94	0.37	0.63	0.002
	Total		374	518	0.42	0.59	<0.0001
2010–11	12/11/10	–	–	–	–	–	–
	01/17/11	A	7	18	0.28	0.72	0.044
2011–12	12/06/11	–	–	–	–	–	–
	01/04/12	A	24	55	0.30	0.70	0.001

A, adult; F, female; M, male; N, nymph; p, test result of comparison of two proportions.

D. maculipennis nymphs were not significant. Significant differences were registered for adults in five sampling moments (Table 1). In February 2008 the sex ratio was biased toward males (1 F:5.25 M) while in early January 2009 there was a bias toward females (1.71 F:1 M). In the three samplings carried out during 2009–10, significant differences were recorded in December 2009 (1 F:1.65 M) and early January 2010 (1 F:1.87 M), where the sex ratio was biased toward males (Table 1). In late January 2010, the proportion of females was higher than males (1.39 H:1 M). Regarding the sex ratio by sampling season (Table 1), significant differences were recorded in only two seasons.

In *B. bruneri*, differences in sex ratio of nymphs were not significant. However, in adults the sex ratio was always significantly different either by sampling time and seasons (Table 2). In all samples, the sex ratio was biased toward males.

Results obtained show that sex proportions recorded in older nymphs for *D. maculipennis* and *B. bruneri* do not deviate from a 1:1 ratio, suggesting that these species, like other grasshopper species, have such a primary sex ratio (Joern and Gaines, 1990). Nevertheless, a significant bias in sex composition in adults of both species was observed. According to several authors biased sex ratios in insect populations could be the result of a variety of causes like the existence of protandry, a differential survival of males and females in response to environmental conditions or natural enemies, different dispersion strategies, or even deficiencies in sampling techniques, among others (Belovsky and Slade, 1993; Sword et al., 2008; Ortego et al., 2011).

The sex ratio in adults of *D. maculipennis* was significantly different in only five of the 18 sampling dates carried out and in two of the seven seasons. In December and early January, time interval at which normally individuals begin to emerge as adults (Mariottini et al., 2011a), significant differences were observed in three sampling moments. Based on these results, it is not possible to detect a trend that would indicate that male adults of *D. maculipennis* emerge first than females. Therefore, protandry seems not to be the case.

A wide range of invertebrate and vertebrate predators, among other natural enemies, affect grasshopper populations (Greathead, 1992; Bardi et al., 2012). Birds are considered the principal vertebrate predator on grasshoppers (Belovsky and Slade, 1993). Some studies suggest that large body size in chemically defended grasshoppers has evolved as a highly beneficial antipredator trait (Whitman and Vincent, 2008). However, there is also evidence that birds tend to select larger body size grasshoppers, modifying the gender composition of the populations (Belovsky and Slave, 1993; Branson, 2005). Gardner and Thompson (1998) reported that female grasshoppers would be preferred by being larger than males. These authors reported that females of *Hesperotettix viridis* (Thomas, 1872) and *Melanoplus aridus* (Scudder, 1878) appeared to be selected more than males by birds. Since *D. maculipennis* females are significantly larger than males (Mariottini et al., 2011b) they could be selected as prey more frequently than males. Although we did not conduct studies on this plausible factor of biased sex ratios, large flocks of a variety of birds were often seen apparently preying in areas of grasshopper aggregations and this could explain the recorded sex ratio biased to males in samplings moments of the 2007–08 seasons and in early January of 2010.

In relation to different sex-related dispersal behavior, it is has been mentioned that males, by being smaller in size, are favored by wind-assisted dispersal, and that females would be more prone to remain close to sites of birth (Sword et al., 2008). For example, Ortego et al. (2011) showed that dispersion in the grasshopper *Mioscirtus wagneri* (Eversmann, 1859) is biased to males, and females tend to be more philopatric. The dispersion behavior of *D. maculipennis* is a key feature in the dynamics of this species and it is an important aspect to consider in future studies. At low densities, *D. maculipennis* appears to show a relatively sedentary behavior, but during outbreak years adults display significant swarm-like displacements reaching distances of up to 50 km (COPR, 1982). In late 2008 and early 2009, an outbreak of *D. maculipennis* of historical magnitude that showed the characteristic *en masse* dispersal flights and affected the study area and several nearby counties was observed (Mariottini et al., 2012). During this period, a sex ratio biased to females was recorded, suggesting the possibility of differential sex-related dispersion in *D. maculipennis*. Our results then could indicate that males have a greater dispersion potential than females.

The sex ratio in adults of *B. bruneri* was significantly different in all samplings, and it was always biased to males. However, the sex ratio recorded in older nymphs was 1:1. These results suggest that just prior to adult emergence or immediately after, a yet unknown factor affected the proportion of females of this species. Mulkern (1983) indicated that temperature, solar radiation, wind speed, and the type and amount of vegetation can also influence the activity of grasshoppers and thus the effectiveness of sampling techniques. This author studied the sex ratio of 11 grasshopper species in the field, and observed that it was constant in each species, which could be attributed to a possible bias in the sampling technique or represent the true composition of populations. We believe, however, that considering the extended sampling period of our study, the number of samples taken, and the amount of individuals collected for the two species, the results presented here would represent the true sex composition of populations of *B. bruneri* and *D. maculipennis* in southern Pampas region.

Conflicts of interest

The authors declare no conflicts of interest.

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